بسم الله الرحمن الرحيم

THE HYPOGLYCEMIC EFFECT OF *CINNAMOMUM VERUM* (ELGERFA) IN RATS

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DEDICATION

I dedicated this study to my mother, my father soul,

My teachers,

My brothers and sisters,

My family,

Friends and to every one hope to learn

With respect

Dalal

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Praise is to the God (Allah) to thank him, on his helping and supporting and sincere appreciation to my supervisor Dr Sulaiman Mohammed El Hassan, Department of Microbiology, Faculty of Veterinary Medicine, University of Khartoum for his valuable guidance, suggestion and sound advance during the study.

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LIST OF ABBREVIATION

ADP	Adenosine diphosphate
ATP	Adenosine triphosphate
dl	Deciliter
DNA	Deoxyribonucleic acid.
DPP-4	Dipeptidyl peptidase-4
EXP	experiment
g	gram
GDM	Gestational diabetes mellitus
GLP	Glucagon-like peptide
GLP-1	Glucagon-like peptide-1
GLP-4	Glucagon-like peptide-4
G.T.T	Glucose tolerance test
h	hour
HDL	High density lipoprotein
IDDM	Insulin dependent diabetes mellitus
Kg	kilogram
LDL	Low density lipoprotein
mg	Milligram
ml	Milliliter
mRNA	Messenger RNA
MHCP	Methylhydroxychalcone polymer
NAD^+	Nicotinamide adenine dinuclutide
NADH	Nicotinamide adenine dinuclutide
NIDDM	Non insulin dependent diabetes mellitus
NO	Nitrogen oxide
nm	Nanometer
P	Probability
POTAGT	Potential abnormality of glucose tolerance. .
RNA	Ribonucleic acid
r pm	Round per minute
SE	Standard error
TZDs	thiazolidinediones

ABSTRACT

This study tested the anti-diabetic and hypoglycemic effect of the Cinnamon which is available in Sudan. Cinnamon is use in so many countries for treatment of bacterial and fungal disease, cold influenza and as antioxidant agent. Recently people discovered that *Cinnamum cassia* induced hypoglycemia in patient with type2 diabetes mellitus. *C.cassia* is closely like *C. verum* (the species found in Sudanese markets). This study was carried out in Wister albino rats.

In experiment (1), the rats were divided into three groups A, B and C; each group consisted of six rats. The serum glucose level of all rats (A, B and C) was measured at 0h after 18 hour fasting then they were injected with glucose solution 50% concentration to induce hyperglycemia. Group B after glucose load was treated orally with Glibenclamide as standard drug. Group C which was also hyperglycemic rats was treated orally with *C. verum* (1g/kgBwt), then the plasma glucose level was measured after 1, 2 and 4h.

In experiment (2): Normoglycemic fasting rats were divided into two groups D and E, each group consisted of six rats. After 18 hours fasting glucose level of both group (D and E) was measured at 0h. Then the rats of group D received orally *C. verum* in dose of 1g/kgBwt while rats of group E were untreated control group. Then serum glucose level of both groups was measured at hour 1, 2 and 4.

Experiment 1 showed that Glibenclamide lowered significantly serum glucose level at hour 1, 2 and 4 when compared to control group

while *C. verum* lowered significantly serum glucose level at hour 2 and nonsignificantly at hour 4.

Experiment (2) showed administration of *C. verum* to normoglycemic rats lowered significantly serum glucose level at hour 4 ($P \leq 0.01$) and nonsignificantly at hour 1 and 2.

This experiment showed that *Cinnamum sp* which found in all market in Sudan has antidiabetic and hypoglycemic effect in hyperglycemic rats. Also the same dose has hypoglycemic effect in normoglycemic rats.

ملخص الاطروحه

في هذه الاطروحه تم دراسة الأثر الخافض لتركيز الجلكوز في الدم والأثر المعالج لمـرض السكرى بواسطة نبات القرفة النوع المتوفر في السودان. ونبات القرفة يستخدم في عدد كثير من دول العالم لعلاج كثير من أمراض الجهاز الهضمي بواسطة الإصابات البكتيريه والفطريـه كمـا يعـالج نزلات البرد وقد اثبت فعاليته كمضاد للاكسدة. وأخيراً أثبتت فاعليته كعلاج لمرض الـسكرى مـن النوع الثاني في الإنسان. والنوع الذي اثبت فعاليته في علاج مرض السكرى هو كثير الشبه بـالنوع المتوفر في السودان. أجريت التجارب في هذا البحث علي الذكور والإناث من جرذان وستر البيضاء.

في التجربة الأولي تم تقسيم الجرذان إلي ثلاثة مجموعات (أ و ب و س) والتي تم حقنهـا بمحلول الجلكوز (بتركيز 50%) بجرعة 2جرام/كيلوجرام من وزن الحيوان لرفع تركيز الجلكوز في الدم. وذلك أثناء إجراء اختبار تحمل الجلكوز وهو بعد إخضاع الفئران للصيام لمدة 18 ساعة ثم اخذ عينات من دمها قبل حقن الجلكوز مباشرة ثم بعد حقن الجلكوز بساعة وساعتين وأربع ساعات لقياس تركيز الجلكوز في الدم. المجموعة (أ) استعملت كمجموعة ضبط ومقارنـة. المجموعـة (ب) بعـد استحداث حالة رفع الجلكوز أخضعت للمعالجة بواسطة دواء مجرب في علاج داء الـسكري وهـو القليبنكلاميد. المجموعة (س) والتي استحدث فيها حالة رفع الجلكوز في الدم ثم أخذت جرعـة مـن القرفة بمقدار 1جرام/كيلوجرام من وزن الحيوان.

في التجربة الثانية تم تقسيم الجرذان إلى مجموعتين (د) و(ى) بعد إخضاعها للصيام لمدة 18 ساعة أعطيت القرفة بالجرعة 1 جرام/1 كيلوجرام من وزن الحيوان. المجموعة (د) والتي لم تخضع لأي معالجات استعملت كمجموعة ضبط و مقارنة.

التجربة الأولي أظهرت أن دواء القليبنكلاميد يخفض الجلكوز في الدم معنويـا فـي الـساعة الأولي والثانية والرابعة عند مقارنته بمجموعة الضبط بينما القرفة تخفض الجلكوز فى الدم معنويا في الساعة الثانية وعدديا في الساعة الرابعة.

التجربة الثانية المجموعة أظهرت أن القرفة تخفض الجلكوز معنويا في الساعة الرابعة وعدديا في الساعة الأولي والثانية .

وهكذا يمكن القول أن القرفة من النوع المتوفر في الأسواق السودانية لها اثر خافض لـسكر الدم في الجرذان ذات المستوى العالى من جلكوز الدم وأيضاً له أثر خافض للجلكوز فى الـدم فـي الجرذان ذات المستوى العادى من جلكوز الدم عندما تعطى بالفم .

INTRODUCTION

People using herbs for a therapeutic purpose in different parts of the world, and this behavior date back to immemorial times. Recently there is a very big attention to medicinal herbs, because of ready availability, their low cost of production and less toxic action compared to those of synthetic compounds (Asima and Pakrashi, 1995).

There are multidisciplinary plant studies, covering: chemistry, pharmacology, biochemistry, microbiology, agential formation and clinical trial. In diabetes mellitus researches, there are so many experiments for using medicinal herbs in treatment of the two types of diabetes, for example using of *Medicago sativa* (Reem, 1999) and *Pimpinella anisum* (Fath ElRahman, 2006). The study of cinnamon and its effect on diabetes mellitus carried out by Khan *et al*. (2003) showed the effect of *Cinnamun cassia* powder with type 2 diabetes. The result of the study showed significant decrease in blood glucose of people with type 2 diabetes.

Objectives:

This study was undertaken to investigate:

a. The effect of oral administration of *Cinnamun verum* barks powder to hyperglycemic rats.
b. The effects of *C. verum* bark powder on normal glycemic rats.
c. To evaluate the effect of *C. verum* as antidiabetic to Glibenclamide as standard drug.

CHAPTER ONE

LITERATURE REVIEW

1.1 Pancreas:

1.1.1 Pancreas gross anatomy:

In man, the pancreas is an elongated organ located across the back of the abdomen, behind the stomach. The right side of the organ (called the head) is the widest part of the organ and lies in the curve of the duodenum (the first section of the small intestine). The tapered left side extends slightly upward (called the body of the pancreas) and end near the spleen (Kumar and Clark, 1988).

1.1.2 Pancreas histological structure:

The pancreas consists of exocrine and endocrine cells. The former are aciner cells, which are grouped into lobules forming the ductal system, which eventually point into the main pancreas duct. The endocrine pancreas consists of hormone-producing cells arranged in nets or islets of langerhans. They do not connect directly to the duct system (Kumar and Clark, 1988).

1.1.3 Pancreatic function:

The pancreas consists of very different organs contained within one structure. The acainer portion of the pancreas has an exocrine function, secreting into the duodenal lumen the enzymes and ions used for the digestive process. The endocrine portion consists of the islets of Langer hans's. The pancreatic islets secret at least four hormones: insulin, glucagons, somatostatin, and pancreatic polypeptide (Robert *et al.*, 1999).

1.2 Insulin:

1.2.1 Structure and secretion:

Insulin is a polypeptide consists of two chains A and B. A chain consists of 21 amino acids, and B chain consists of 30 amino acids (Fig. 1). The two chains of A and B are linked by two intrachain disulfide bridges connect residues 6 and 11 of the A chain. Insulin is synthesized as preprohormone (molecular weight approximately 11,500), modified to proinsulin 9000-molecular weight and then to its mature structure, this is done by specific enzymatic cleavage. The proinsulin molecule is transported to the Golgi apparatus wherein proteolysis and packaging into the secretary granules begin. The human pancreas secretes 40-50 units of insulin daily, which represent about 15-20% of the hormone stored in the gland. Insulin secretion is an energy requiring process that involves the microtubules microfilaments system in the B-cell of the islets (Robert *et al*., 1999).

1.2.2 Factors affecting insulin release:

1.2.2.1 Glucose:

The maximal response is obtained when glucose levels are between 300 and500 mg/dl. It is generally accepted that an increase of the ATP/ADP ratio result in the inhibition of ATP-sensitive potassium ion efflux channels. This causes depolarization of the B-cells and activation of voltage-sensitive calcium ion channels. The calcium ion influxes result in insulin secretion (Robert *et al*., 1999).

Fig. 1: Insulin structure, A and B chains linked by disulphide bonds.

1.2.2.2 Hormonal factor:

Numerous hormones affect insulin release, like: α -adrenergic agonist, B-adrenergic agonist, growth hormone, cortisol, placenta lactogen, estrogen and progestron.

1.2.2.2.1 Probable actions of hormones countering the effect of insulin in man:

During an oral tolerance test, a mixed meal the first one-and-a-half hours are dominated by increased secretion of insulin, and both growth hormone and glucagon secretion are inhibited. Cortisol and adrenal hormones levels did not change significantly.

Exercise represents a special stress with a rapid increase in the demand for metabolic fuel. At rest 90 % of the energy requirements of muscle come from oxidation of stored glycogen but, if the demand for oxygen outstrips supply, anaerobic glycolysis becomes all important. Glycogen supplies are rapidly depleted and glucose is extracted from the circulation independent of insulin. Blood glucose levels fall and secretion of insulin also decreases. Catecholamines and cortisol level rise, stimulating lipolysis and gluconeogenesis. The increase in hepatic glucose production matches the increased extrahepatic utilization so that glucose levels do not change markedly (Edward *et al.,* 1995).

1.2.2.3 Pharmacological agents:

Many drugs stimulate insulin secretion, but the sulfonylurea compounds are used most frequently for therapy in human. Drugs such as tolbutamide stimulate insulin, release by a mechanism different from that employed by glucose and have achieved widespread use in the treatment of non insulin – dependent diabetes mellitus (type 1). A receptor that

binds this class of drugs has recently been cloned from the pancreatic B-cell. This receptor is closely linked to the ATP-sensitive potassium ion channels described above, which may explain the mechanism of action of this important class of drugs (Robert *et al.,* 1999).

1.2.3 Insulin function:

Insulin action includes membrane transport of glucose, amino acid and certain ions; formation of triglyceride; increase storage of glycogen; stimulation of DNA, RNA and protein synthesis.

1.2.3.1 The mechanism of insulin action:

Insulin action begins when the hormone binds to specific glycoprotein receptor on the surface of the target cell. The diverse action of the hormone (Fig. 2) can occur within seconds or minutes. The insulin receptor is a hetrodimer consisting of two subunit, designated α and B, in the configure action α 2 B2, linked by disulfide bonds. Both subunits are extensively glycosylated, and removal of sialic acid and galactose decreases insulin binding and insulin action. Each of these glycoprotein subunits has a unique structure and function. The A subunit (5KDa) extracellular, and it bind insulin, probably via a cysteine-rich domain. The B subunit (95KDa) is transmembrane protein that performs the second major function of receptor. The cytoplasmic protein of the B subunit has tyrosine kinase activity and an autophosphorylation site (Robert *et al.,* 1999). Recently the work from several laboratories described the cellular life cycle of the IR, shown schematically in Fig. (2) in five cellular compartments (colored blocks).

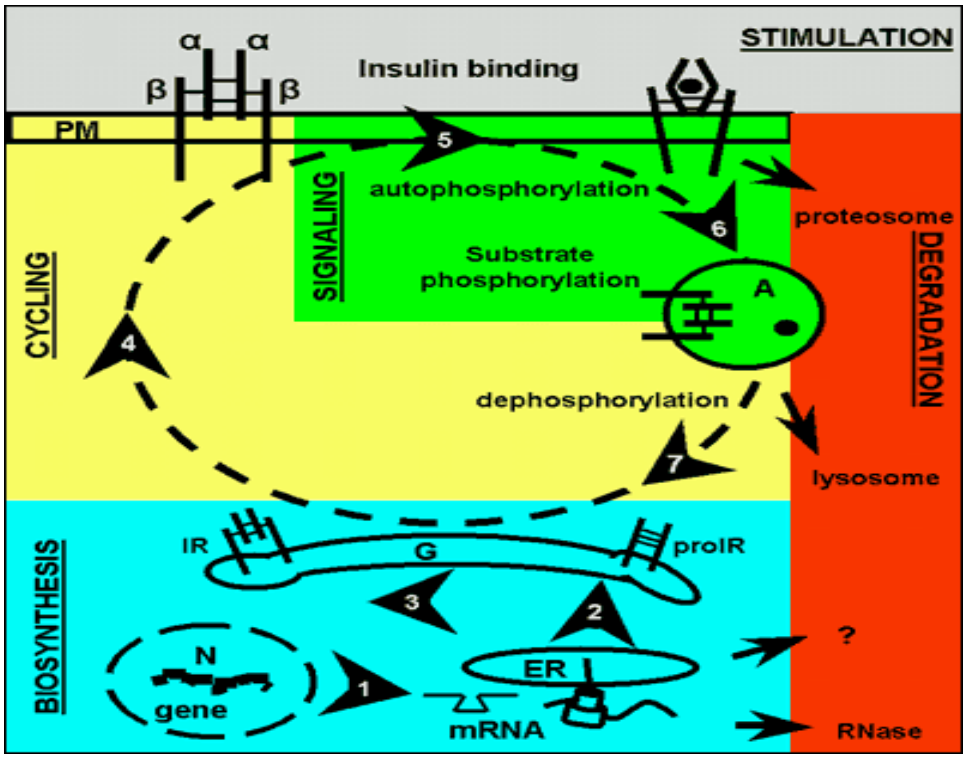

Fig. 2: Insulin binding its receptor and the mechanism of action. Life cycle of the insulin receptor, it is shown as four parallel lines.

Receptor internalization is also triggered by insulin binding. Insulin is removed from circulation by the IR, through endocytosis (Terris *et al*., 1979).

1.3 diabetes mellitus:

Diabetes mellitus is a syndrome characterized by chronic hyperglycemia that results from insulin deficiency or resistance or both (Kumar and Clark, 1988).

1.3.1 Diabetes mellitus in domestic animals:

Diabetes mellitus is frequently found in the dog (1 in 152), and cats (1 in 800), although it has been reported in horse, cattle, sheep and pigs. The fundamental defect in domestic animals is an absolute or relative lack of insulin. As a result of this deficiency in insulin, the animal is unable to utilize glucose. Consequently hyperglycemia is constant finding in diabetic animals. Clinical signs typically associated with diabetes include polydipsia, polyurea, increase appetite and weight loss. The classic signs of ketosis include anorexia; vomiting, weakness, lethargy and increase respiratory rate (Coles, 1986).

1.3.2 Diabetes mellitus in man:

Diabetes mellitus affects more than 30 million people world wide (Kumar and Clark, 1988). In 1993 there were 7.8 million diagnosed cases of diabetes in the United State, of whom 43 % were treated with insulin. Insulin dependent diabetes mellitus (IDDM) with onset at age < 30 years comprises 7% of all diagnosed cases.

Some studies indicate that 7 % of insulin-treated cases with onset at age ≥ 30 years may also be IDDM. Impaired glucose tolerance (IGT) is a class that encompasses persons whose glucose tolerance is intermediate between normal and diabetic. About 11% of adults have IGT when tested by oral glucose challenge (Michael *et al.,* 1997).

1.3.3 Classification of diabetes mellitus:

Most patients can be classified clinically as having either insulin-dependent diabetes mellitus (IDDM or type1) or non insulin-diabetes (NIDDM or type11). In 1979, the national diabetes data group formally classified diabetes mellitus and other categories of glucose intolerance as follows:-

1.3.3.1 1nsulin dependent diabetes mellitus (IDDM or type1):

About 10% of diabetic patients are insulin dependent diabetes mellitus.Type1 diabetes is a usually results when the body's own immune system launches a misguided attack on the insulin-producing beta cells in the pancreas. Harmful immune system cells, including some T cells, are normally eliminated during their maturation. However, in susceptible individuals, this disease – causing T cells initiate an inflammatory process in the pancreas that eventually leads to the destruction of beta cells. The other arms of immune system – the B cells – produce antibodies that also recognize beta cell protein (Dorman et *al.,* 1995).

Type 1diabetes may develop in persons with a family history of type 1 diabetes, but may also develop in persons with no family history of diabetes. In either case, the person has one or more genes that make

them susceptible to the disease. Environmental factors, such as exposure to certain viruses and foods early in life, may trigger the autoimmune response (Edward *et al*., 1995).

1.3.3.2 Non-insulin-dependent diabetes mellitus (NIDDM or type2):

About 90% of persons with diabetes are non-insulin dependent diabetes mellitus (type 2) patients, such patients are usually obese, have elevated plasma insulin level, and have down regulated insulin receptors (Robert *et al.,* 1999).The pathogenesity of type2 diabetes is complex and involves the interaction of genetic and environmental factors (Henry *et al.,* 2007). Environmental factors, genetic factors, age, pancreatic pathology and pregnancy may contribute in the development of type 2 diabetes mellitus.

1.3.3.2.1 Environmental factors:

Particularly excessive caloric intake and sedentary lifestyle lead to obesity. The clinical presentation is also heterogeneous with a wide range in age of onset. Severity associated with hyperglycemia, and degree of obesity. From a pathophysiologic standpoint, person with type2 diabetes constantly showed three cardinal abnormalities: (1) resistance to the action of insulin in peripheral tissue, particularly muscle and adipose tissue but also liver; (2) defective insulin secretion, particularly in response to glucose stimulus; and (3) increase glucose production by the liver.

1.3.3.2.2 Genetic factor in the developing type2 diabetes:

Generally type 2 diabetes consists of monogenic and polygenic forms. The monogenic form is uncommon disorder.

I.3.3.2.2.1 Monogenic forms of diabetes associated with insulin resistance:

More than 70% mutations have been identified in the insulin-resistance patients. These mutations may impair receptor function by a number of different mechanisms, including decreasing the number of receptors expressed on the cell surface e.g. by decreasing the rate of receptor biosynthesis (class1), accelerating the rate of receptor degradation (class5), or inhibiting the transport of receptors to the plasma membrane (class2). The intrinsic function of the receptor may be abnormal if the affinity of insulin binding is reduced (class3) or receptor tyrosine kinase is inactivated (class4). The insulin resistance that is associated with insulin receptor mutations may be severe and present in the neonatal period, as with leprechaunism and Rabson Mendenhall syndrome, or occur in a milder form in adulthood leading to insulin-resistant diabetes with marker hyperinsulinemia, acanthosis nigricans, and hyperandrogenism (Henry *et al*., 2007).

1.3.3.2.3 Age:

In Britain over 70% of all cases of diabetes, occur after the age of 50 year. In contrast IDDM which mainly affects younger people, NIDDM is principally a disease of the middle –aged and elderly. Thus, aging is an important risk factor for NIDDM (Edward *et al*., 1995*)*.

1.3.3.2.4 Pregnancy:

During normal pregnancy, the level of plasma insulin is raised by the action of placental hormones, thus placing a burden in insulin-secreting cells of the pancreatic islet. The pancreas may be unable to

meet these demands in women genetically predisposed to develop both types of diabetes. Repeated pregnancy may increase the likelihood of developing permanent diabetes, particularly in obese women. Long-term studies showed that some 80% of women with gestational diabetes ultimately develop permanent clinical diabetes requiring treatment (Edward *et al.*, 1995).

1.3.3.2.5 Pancreatic pathology and insulin secretion:

In contrast to IDDM where at diagnosis the insulin- secreting cells have largely disappeared from the pancreas so that plasma immunoreactive insulin is either very low or undetectable, in NIDDM there is only moderate reduction in the total mass of islet tissue consistent with a measurable, though, when related to the blood glucose level, and reduced concentration of insulin in plasma. There are, however, some pathological changes, which are typical of NIDDM and demonstrable in most, although not all cases. The most consistent of these changes is probably deposition of amyloid which is accompanied by atrophy of the normal tissue, particularly islet epithelial cells. In more advanced lesion, the islet is more or less converted to amyloid and the reduction in the number of insulin-secreting cells is more pronounced than that of glucagon-secreting-cells. Deposition of amyloid is probably not a cause of diabetes but rather reflects a pathological process which is increased in NIDDM (Edward *et al.*, 1995).

1.3.3.2.6 Insulin resistance and the risk of type2 diabetes:

The term insulin resistance indicates the presence of an impaired biologic response to either exogenously administered or endogenously

secreted insulin. Insulin resistance is manifested by decreased insulin-stimulation glucose transport and metabolism in adipocytes and skeletal muscle and by impaired suppression of hepatic glucose out put. Insulin sensitivity is influenced by a number of factors including age, weight, ethnicity, body fat (especially abdominal), physical activity, and medication (Henry, .2007).

1.3.3.2.7 Obesity and type2 diabetes:

The absolute amount of body fat has an effect on insulin sensitivity across a broad range. Elevated free fatty acids (FFAs) predict the progression from impair glucose tolerance test to diabetes. In the periphery, free fatty acids may not be markedly elevated because of efficient extraction by the liver and skeletal muscle. Increases in fatty acid flux to skeletal muscle related to the increase visceral lipolysis have been implicated in the inhibition of muscle glucose up take. The Randle hypothesis, or glucose fatty acid cycle, was originally proposed to account for the ability of free fatty acids to inhibit muscle glucose utilization. Randle and co-workers (1966) demonstrated that fatty acids compete with glucose for substrate oxidation in isolated muscle. The increase in fatty acid metabolism leads to an increase in the intra mitochondrial acetyl Co enzyme A (Co A); Co A and reduced nicotinamide, NADH/NAD ratios with subsequent inhibition of puruvate dehydrogenase. The resulting increased intercellular mitochondrial (and cytosolic) citrate concentration result in allosteric inhibition of phosphofructokinase, the key rate controlling enzyme in glycolysis. The subsequent accumulation of glucose 6 phosphate would inhibit

hexokinase II activity, resulting in an increase in intercellular glucose concentrations and decrease glucose up take (Michael *et al.,* 1997).

1.3.4 Intermediate risk categories:

1.3.4.1 Impaired glucose tolerance (IGT):

Impaired glucose tolerance (IGT) is defined as glycemic response to the standard 75g oral glucose challenge that is intermediate between normal and diabetic i.e. venous plasma or capillary whole blood glucose concentration 140-199mg/dl at 2 hour after a glucose load. The incidence of the IGT was reported to be 17.6 per 1,000 person-years in non Hispanic whites and 32.6 per 1,000 person- years in Hispanic. The importance of the IGT as a prognostic category for development of NIDDM is generally accepted, but some aspects remain controversial (Michael *et al.,* 1997).

1.3.4.2 Potential abnormality of glucose tolerance (Pot AGT):

Person who have never exhibited abnormal glucose tolerance but who are at substantially increased risk for the development of IDDM and NIDDM are well established, such as being a relative of an IDDM or NIDDM diabetic or belonging to certain ethnic or racial group, although the degree of risk for any of the specific circumstances is much less clear.

1.3.5. Biochemical changes associated with diabetes mellitus:

Lack of insulin causes significant disturbance of carbohydrate, protein and fat metabolism (Kumar and Clark, 1988).

1.3.5.1 Disturbance in carbohydrate metabolism:

The disturbance in carbohydrate metabolism is due to the fact that the liver and skeletal muscles cannot store glycogen and the muscles

are unable to utilize glucose. When the kidney threshold for glucose is exceeded, glucouria occurs with consequent increase of water excretion and disturbance of electrolyte and water balance.

1.3.5.2 Disturbance in protein metabolism:

Protein metabolism in the liver is also deranged and an excessive amount of protein is transformed into carbohydrate (Wilson *et al.,* 1975).

1.3.5.3 Disturbance in fat metabolism:

In addition, the amount of fat metabolized by the diabetic patient is excessive, and since normal fat catabolism can only proceeds at a limited rate, ketone bodies are present in the blood and the urine in much large amount than normally; these substances are excreted in the urine as β-hydroxybutaric acid and acetoacetic acid and as acetone in the breath (Wilson *et al.*,1975).The liver produces ketone bodies by oxidation of the free fatty acids and this decreases the production of ketone bodies, conversely, glucagons stimulates ketone body production by stimulating lipolysis and increasing fatty acid oxidation. In the diabetes patient, particularly with type1 diabetes, the consequences of insulin deficiency and glucagons excess provide a hormonal milien that favors ketogenesis and in the absence of appropriate treatment, may lead to ketoneamia and acidosis. Insulin also interacts with the capillary endothelium to activate lipoprotein lipase, the enzyme that hydrolyses the triglycerides present in very-low-density lipoprotein VLDL and chylomicrons, resulting in release of intermediate-density lipoprotein ILD particles (Taskinen, 1985). These ILD particles are converted by the liver to the more cholesterol- rich- density LDL. In the untreated or under treated diabetic

patient, hypertriglycridemia may occur as a result of the decrease removal of VLDL secondary to decrease activity of lipoprotein lipase. In addition, deficiency of insulin may be associated with increased production of VLDL (Gilman *et al.,* 1992).

1.3.5.4 Pathological complication of diabetes mellitus type 2:

Type 2 may go unnoticed for years in a patient before diagnosis, since the symptoms are typically milder (no ketoacidosis) and can be sporadic. However, severe complications can result from improperly managed Type 2 diabetes. The case of diabetic complication is not known and may be multifactorial. Major emphasis has been placed to sorbitol by the enzyme adol reductase. Sorbitol which appears to function as a tissue toxin, has been implicated in the pathogenesis of retinopathy, neuropathy, cataracts, nephropathy and artic disease. The mechanism is perhaps best worked out in experimental diabetic neuropathy, where sorbitol accumulation is associated with a decrease in myoinsitol content, abnormal phosphoinositide metabolism, and decrease in Na, K ATPase activity.

Second mechanism of potential pathogenic importance is glycation for non enzymatic addition of hexoses to proteins and glycosylation for enzymatic addition. The effect of such glycation on hemoglobin is important in monitoring the control, but multiple proteins in the body are altered in the same way, often with disturbed function. Examples include plasma albumin, fibrin, collagen, lipoproteins and the glycoprotein recognition system of hepatic endothelial cell (Harrison, 1996).

1.3.6. Therapy for diabetes:

The most recent information reported the use of diet, oral agent, and insulin by people with diabetes mellitus (Michael *et al.*, 1997).

1.3.6.1 Nutritional therapy:

Nutritional therapy is a challenging but necessary dimension in the management for all types of diabetes. For children with IDDM, a goal is to match diet with insulin requirements to ensure normal growth and development. According to current guidelines, dietary protein intake should constitute 10-20% of total daily calories. Saturated and polyunsaturated fat should each be limited to <10% of total daily calories and the remaining 60-70% of calories, composed of monounsaturated fat and carbohydrate. Cholesterol should be limited to ≤ 300 mg daily (Michael *et al*., 1997).

1.3.6.2 Pharmacological treatment:

There are so many drugs in treatment of type 2 diabetes like:-

1.3.6.2.1 Meglitinides:

Meglitinides help the pancreas to produce insulin and are often called "short-acting secretagogues." Their mode of action is original, affecting potassium channels (Rendell, 2004). By closing the potassium channels of the pancreatic beta cells, they open the calcium channels, hence enhancing insulin secretion (Eurich *et al*., 2007).

1.3.6.2.2 Peptide analogs:

Incretin mimetics:

Incretins are insulin secretagogues. The two main candidate molecules that fulfill criteria for being an incretin are Glucagon-like

peptide-1 (GLP-1) and Gastric inhibitory peptide (aka glucose-dependent Insulinotropic peptide or GIP). Both GLP-1 and GIP are rapidly inactivated by the enzyme dipeptidyl peptidase-4 (DPP-4) (Thorens, 1995).

1.3.6.2.3 Glucagon-like peptide (GLP) analogs:

GLP agonists bind to a membrane GLP receptor as a consequence of this, insulin release from the pancreatic beta cells is increased. Endogenous GLP has a half life of only a few minutes; thus an analogue of GLP would not be practical (Eurich *et al*., 2007).

1.3.6.2.4 Thiazolidinediones:

Thiazolidinediones (TZDs), also known as "glitazones," bind to PPARγ, a type of nuclear regulatory proteins involved in transcription of genes regulating glucose and fat metabolism. These PPARs act on Peroxysome Proliferator Responsive Elements (PPRE). The PPREs influence insulin sensitive genes, which enhance production of mRNAs of insulin dependent enzymes. The final result is better use of glucose by the cells (Eurich *et al.,* 2007).

1.3.6.2.5 Glibenclamide:

Also known as glyburide, is an anti-diabetic drug in a class of medications known as_sulfonylureas, used in the treatment of type II diabetes, it is one of only two oral anti-diabetics in the World Health Organization Model List of Essential Medicines (the other being metformin). It is also sold in combination with metformin under the trade name (Marcha, 2007).

1.3.6.2.6 Glucovance:

Glucovance is a drug works by inhibiting ATP-sensitive potassium channels in pancreatic beta cells. This inhibition causes cell membrane depolarization, opening of voltage-dependent calcium channels, thus triggering an increase in intracellular calcium into the beta cell which stimulates insulin release(Monami *et al*., 2006).

1.4 Medicinal plants:

Because of the ready availability of the medicinal herbs, their low cost of production and less toxic action compared to those synthetic compounds, the World Health Organization has been advocating the use of traditional remedies. Multidisciplinary plant studies, covering chemistry, pharmacology, biochemistry, galenical formulation, standardization, clinical trials and production technology have resulted in technical processes being developed for production, semi finished products and medicaments from indigenous medical plants. Phytochemical studies, one of the advances in research and application of modern technical method, which determined the chemical composition of medicinal plants so as to identify the active chemical ingredient (Asima and Pakrashi, 1995).

Recently, many experimental and clinical trials were made to detect the hypoglycemic and anti-diabetic effects of many medical plants used in the medicine for the treatment of diabetes mellitus (FathElrahman, 2006).

1.4.1 *Cinnamomum verum*:

Cinnamon considered more precious than gold, in the recent studies in medical herbs. During this time, cinnamon received attention in china, evidenced by its inclusion in one of the earliest books on Chinese botanical medicine, dated around 2700B.C. even today, Cinnamon is considered one of 50 Chinese fundamental medicinal herbs (WHFoods, 2007).

1.4.1.1 Classification of *Cinnamomum verum*:

Kingdom: Plantae.
Division: Magnoliphyta.
Class: Magnolipsida.
Order: Laurales.
Family: Lauracae.
Genus: Cinnamomum.
Species: *Cinnamomum verum* J.s.Persl.
Synomons names: *Cinnamomum zeylanicum.*
English names:-Cinnamon, Ceylon Cinnamon, Chinese cinnamon.
Arabic name: Algerfa.

1.4.1.2 Plant origin:

The member of the family Lauracae can be found throughout tropical and subtropical regions of the world, Primary in the lowland to mountains rain forests. The main centers are in Southeast Asia and tropical America. Lauracae family includes about 32 genera, and 2500 species (Heywood *et al.,* 1971). *Cinnamomum cassia* (Fig. 4) was founded in China, species closely related to *Cinnamomum verum*

1.4.1.3 Plant description:

Cinnamon, *Cinnamomum* spp, is an aromatic spice isolated from the inner bark of cinnamon trees (Fig. 3). Which is a green tree all the seasons of the year, its height about 12 miters and it has horizontal branches, simple and ovate leaves, yellow or white flowers (Fig.4, 5) and its fruit is globe or ovate shaped coumarin (Elgazali *et al*., 1998).

1.4.1.4.1 Active ingredients of *Cinnamomum verum*:

Cinnamomum spp. bark contains: between 0.5-4% essential oil containing 80% cinnamaldehyde, up to 10% eugenol and 5-10% trans-cinnamic acid; 4-10% phenolic compounds; condensed tannins; catechins; oligomeric proanthocyanidins; other monoterpenes including limonene and alpha-terpinol; sesquuiterpenoid including pinene; calcium monoterpenoid oxalate; gum; mucilage; resin; starch; sugar; and traces of coumarin (Mancini *et al*.,1998).

1.4.1.5 Biological activities:

1.4.1.5.1 Antimicrobial effect of cinnamon:

The antimicrobial effect of *Cinnamomum* sp was identified in a laboratory experiment in which pure *Cinnamomum cassia* extract, mainly composed of the active ingredient cinnamaldehyde, was tested on isolated strains of bacteria, including Gram-positive *Staphylococcus aurus*, gram-negative *Escherichia coli*, *Enterobacte*r *aerogenes, Proteus vulgaris, Pseudomonas aeruginosa*, *Vibrio cholerae*, *Vibrio parahaemolyticuas*, and *Samonella typhymurium*.

The antimicrobial effect resulted in a minimum inhibition concentration (MIC) of *Cinnamomum cassia* extract ranging from 75 mcg/mL to 600 mcg/mL on these various bacteria (Ooi *et al.*, .2006).

Fig. 3*: Cinnamomum verum* bark

Fig. 4*: Cinnamomum cassia* (flowering plants)

Fig. 5*: Cinnamomum verum* (flowering plants)

However, conflicting data was obtained in a randomized, controlled pilot clinical trial of 15 patients ages 16 to 79 who had a positive Campylobacter urease test for *Helicobacter pylori*. Each patient in the control group received 40 mg of ethanol extract of cinnamon twice daily for four weeks; the control group received plain 95% ethanol. With urea breath tests as the measurement of efficacy, the mean urea breath test before and after the study for the treated group were 22.1 and 24.4, respectively; the mean urea breath test before and after the study for the controlled group were 23.9 and 25.9, respectively. This conclusive study found that 40 mg of cinnamon extract given twice dialy was ineffective in eradicating *Helicobacter pylori* (WHFoods, 2007; Nir *et al.*, 2000).

1.4.1.5.2 Anti-inflammatory:

Although Cinnamon historically had not been used to treat inflammatory disorders, its anti-inflammatory effect was demonstrated experimentally. Specifically, *Cinnamomum cassia* was used to investigate the anti-inflammatory effect on nitric oxide (NO) and nuclear factor kappa-b. Both substances have been implicated in inflammation. In acute and chronic inflammation, there is an increased production of NO, which promotes vasodilatation and results in increased vascular permeability and edema. Nitric oxide also activates COX-2 enzyme involving in the biosynthetic pathway of inflammatory prostaglandins (Thomas *et al.*, 2004). Nuclear factor kappa-b contributes to inflammation through induction of transcription of genes coding for inflammatory mediators. It was found that cinnamaldehyde, specifically 2'-hydroxycinnamaldehyde found in *Cinnamomum cassia*

extract, exhibited a dose-dependent inhibitory effect on NO production and transcriptional activity of NF-kB, thereby contributing to its anti-inflammatory qualities (Lee *et al.,* 2005).

1.4.1.5.3 Antifungal:

Cinnamon oil has been reported as an antifungal agent, although current efficacy of cinnamon oil's fungicidal effect has not been tested in clinical trials.

Experimentally, promising results on its antifungal activity were reported in two *invitro* studies of cinnamon oil on *Cryptococcus neoformans* and *Aspergillus niger*. *Cryptococcus neoformans* is an opportunistic fungal pathogen affecting the lungs or meninges of immunocompromised or AIDS patients, causing pulmonary cryptococcosis *meningitis* (Lee *et al*., 2005). It was found that the phenolic compound in cinnamon oil identified as eugenol is responsible for its fungitoxic activity (Viollon and Chaumont, 1994). Cinnamon oil's antifungal property was again demonstrated in a more recent in vitro study on *Aspergillus niger (A. niger),* an opportunistic fungal pathogen residing in the air and, through inhalation of *Aspergillus sp*. spores, entering the respiratory tract of patients with AIDS or with immunocrompromised conditions to cause Aspergillosis (Beers and Porter, 2006). Seventy- *Cinnamomum cassia*, were tested for the inhibition of five botanical essential oils, including *Cinnamomum zeylanicum* and hyphal growth and spore formation on inoculated agar with *A. niger* incubated at 28^{o}C for 48 hours. Among the 75 botanical essential oils used, *Cinnamomum zeylanicum* and *Cinnamomum cassia*

demonstrated maximal and superior results; the zone of hyphal growth inhibition and zone of spore formation were 43 and 40 versus 50 and 45 for *Cinnamomum zeylanicum* and *Cinnamomum cassia*, respectively (Pawar and Thaker, 2006).

1.4.1.5.4 Antioxidant:

There is more to cinnamon besides the antimicrobial, anti-inflammatory, and antifungal effects attributed to cinnamaldehyde found in *Cinnamomum cassia* powder and oil. The presence of oligomeric proanthocyanidins (OPC), a class of bioflavonoid, opened a new area of research on its antioxidative effect. Through agriculture research, type A and type B oligomeric proanthocyanidins were identified in cinnamon spice via mass spectrometer analysis. Additionally, it was found that over 84% to 90% of OPC found in cinnamon spice were type A OPC (Gu *et al*., 2003). However, there has not been a specific study on the antioxidative effect of cinnamon alone. Antioxidants are essential to the human body to neutralize free-reactive oxygen species, also known as free radicals, to maintain functional cellular membrane and structure. Furthermore, free radicals associated with impaired glucose metabolism and antioxidants have been implicated in the regression of diabetes mellitus.

1.4.1.5.5 Hypoglycemic effect of *Cinnamomum* sp:

The quest for new treatments continues as the realm of research for type 2 diabetes expands to nutraceutical products. A pilot clinical trial conducted in Pakistan to study the effect of cinnamon in patients with type 2 diabetes harvested successful results. The study involved 60

subjects (30 men and 30 women) with type 2 diabetes, average age of 52.2±6.32 years, fasting plasma glucose of 140-400 mg/dL, and not taking insulin or medicines for other health conditions. Subjects were randomly assigned into three placebo groups and three cinnamon groups in which they were given 1, 3, or 6 g of cinnamon capsules (*Cinnamomum cassia* powder) daily for 40 days followed by a 20-day wash-out period. All subjects were allowed to continue taking sulfonylurea drugs during the study. Plasma glucose, triglyceride, low-density lipoprotein (LDL), high-density lipoprotein (HDL), and total cholesterol were measured at fasting state before and after treatment. Effects were similar among the three cinnamon-treated groups. The mean reduction in blood glucose, triglyceride, LDL, and total cholesterol levels were 18–29, 23–30, 7–27, and 12–26%, respectively; changes in these levels were not significant in the placebo groups as well as HDL levels for all six groups. Interestingly, the effects of cinnamon lingered on after discontinuation. Khan *et al.* (2003). reported that plasma glucose, triglyceride, LDL, and total cholesterol levels continued to remain lower than baseline during the 20-day wash-out period. Khan *et al*. (2003) suggested that daily consumption of cinnamon may not be necessary due to the observed sustained effects of cinnamon in studied subjects with type 2 diabetes and that inclusion of cinnamon into daily diet may be beneficial to the remainder of the population.

CHAPTER TWO
MATERIALS AND METHODS

This work has been conducted in the Department of Pharmacology in Medicinal and Aromatic Plants Institute (MAARI), Sudan National Center for Research.

2.1 Material

2.1.1 Experimental Animals

Charles Foster strain white albino rats divided to five groups, each group consisted of six rats, their body weight between (150-200g), were used for all experiments. Rats used in all experiments were kept on a fixed diet so as to stabilize the fasting serum glucose.

2.1.2 Feeding

Each group was kept in a cage and was supplied with feed composed of meat, wheat flour, carrot and salt. The feed was available at the rate of 36 g per day in each cage.

2.1.2.1 Chemical analysis of the rat diet

- Dry matter 97.22%
- Ash 3.82%
- Crude protein 7%
- Crude fibre 5.61%
- Nitrogen free extract 73.21%
- Fat 7.52%

The analysis of the diet was made at the Department of Animal Nutrition, Faculty of Animal Production, U. of K., Sudan.

2.1.3 Plant material:

The *Cinnamomum verum* bark was bought from Khartoum Bahry market. Dry bark of Cinnamon was ground to powder.

2.2.3 Experimental protocol:

Experimental design of experiment 1 is shown in table 1and experimental design of experiment 2 is shown in table 2.

Experiment 1:

2.1.3.1. Induction of hyperglycemia in rats:

Albino rats in group A, B and C were kept fasting for 18 hours. The serum glucose level was determined at zero hour. Immediately after that 50% of glucose solution was injected interaprotonially at dose of 2g/kg Bwt so as to induce hyperglycemia.

2.1.3.2. The effect of oral administration of *Cinnamomum verum* bark on serum glucose level of hyperglycemic rats:

Group A:

After induction of hyperglycemia, the serum glucose level of the animals was measured at 1 hour, 2hour and 4 hour after glucose load.

Group B:

After induction of hyperglycemia, Glibenclamide, standard antidiabetic drug was given per os at a dose of 10mg/kgBwt. The glucose was then measured at 1hour, 2hour and 4 hour.

Group C:

After inductions of hyperglycemia, cinnamon bark powder mixed with distilled water was given orally at a dose of 1g/kg Bwt. The plasma glucose level of the animals was measured at 1 h, 2h and 4 h.

Table No 1: The experimental design of experiment (1).

Description	Groups		
	A	B	C
Numbers of rats	6	6	6
Induction of hyperglycemia	+	+	+
Treatment	-	Glibenclamide	*C. verum*

Table No 2: The experimental design of experiment (2).

Description	Group	
	D	E
Number of rats	6	6
Induction of hyperglycemia	-	-
Treatment	-	*C. verum*

Experiment2:

2.1.3.4 The effect of the oral administration of *Cinnamomum verum* bark powder on serum glucose level of normoglycemic rats:

Group D:

Group D was kept fasting for 18 hours. The glucose was then measured at zero, 1, 2 and 4h after fasting.

Group E:

Albino rats of group E were kept fasting 18 hours. The serum glucose level was measured at zero h, immediately after that *Cinnamomum verum* bark powder mixed with distilled water was given orally at a dose of 1g/kg Bwt. The plasma glucose level of the animals was then measured at 1, 2, and 4 h following treatment.

2.1.4 Sample collection:

In all groups, blood was drawn out by capillary tube in sterile test tubes from the orbital plexus of rats under inhalation anesthesia using halothane according to Khana, (1992). The collected blood was allowed to clot and then centrifuged at 300 r p m for 5 minuets to separate serum. The prepared serum was used to estimate, glucose.

2.1.5 Chemicals and reagents:

Glucose's kit of Crescent Diagnostic laboratories was used.

2.1.6 Instruments:

* IEC-CENTRA-4B centrifuge.
* Finn pipettes, digital micropipette was used for pipetting serum samples and reagent.
* Sterile plain test tubes, was used for collection of blood.

* CIBA CORNING colorimeter model 252 were used to estimate glucose.

2.2 Methods:

2.2.1 Glucose oxidase method: GOD/PAP:

Glucose concentration in the serum was determined by enzymatic colourimetric, GOD-PAP method (Trinde, 1969).

Principle:

Glucose is determined after enzymatic oxidation in the presence of glucose oxidase. The hydrogen peroxide formed reacts under catalysis of peroxidase, with phenol and 4-aminophenazone to form a red violet quinoneimine dye as indicator.

Reaction principle:

1. α-D-Glucose $\xrightarrow{\text{mutarotase}}$ β-D glucose.
2. β-D glucose+ $H_2O + O_2 \xrightarrow{\text{GOD}}$ D-gluconic acid + H_2O_2.
3. H_2O_2 + 4 aminophenazone + phenol $\xrightarrow{\text{POD}}$ quinonemine + 4 H_2O.

Reagent composition:

Crescent Diagnostic, reagent (Cat No.CS605s) was used. It is composed of:

1. Phosphate buffer (pH 7.5) 0.1 mol/l

* Aminophenazone 0.25 mmol/l
* Phenol 0.75 mmol/l
* Glucose oxidase 15 KU/l
* Peroxidase 1.5 KU/l
* Mutarotase 2 KU/l

2. Glucose standard 100mg/dl or 5.5mmol/l

Procedure:

A volume of 0.01µl of each standard, sample and distilled water was pipitted each in separate cuvettes, 1µl of the reagent was added to each cuvettes, mixed well and incubated for 10 minutes at 20-25^{o}C or 37^{o}C. The absorbance of the standard A_{std} and of the sample A_s were measured against the reagent blank at the wave length 546-500 nm.

Calculations:

$$\text{Glucose (mg/dl)} = \frac{A_s}{A_{std}} \text{ X concentration of standard}$$

2.2.4 Statistical design:

According to completely randomized design, the rats of experiment 1 and 2 were divided into three groups and two groups respectively. Each group contains 6 rats of similar body weight. The data were analyzed by statistical package for social scientists (SPSS) and T-test. Each test was conducted at 5% level of significance.

CHAPTER THREE

RESULTS

Experiment 1

3.1 The effect of oral administration of *Cinnamomum verum* and Glibenclamide on serum glucose level of hyperglycemic rats

When *C. verum* and Glibenclamide were administrated orally to fasting rats of group B and C, the serum glucose level measured after 18 hours fasting and then at 1, 2 and 4 hours after glucose loading are shown in Table (3) and Fig. (6).

At 0 h, there was no significant difference in glucose level between the treated groups (B and C) and the control group (A) as shown in Table (3) and Fig. (6).

At hour 1after glucose loading, the serum glucose level of Glibenclamide treated rats group (B) was significantly ($P \leq 0.0007$) lower than control group (A) and *C. verum* treated group (C). But, the serum glucose level of *C. verum* treated group (C) was nonsignificantly higher than that of control group (242.08 Vs 208.58), as shown in Table 3 and Fig. 6.

At 2h after glucose loading the glucose level of both treated group (B and C) were significantly ($P \leq 0.001$) than that of control (Table 3, Fig. 6) and the glucose level of Glibenclamide treated group was nonsignificantly lower than that of *C. verum* treated group C.

When serum glucose was measured at hour 4, the glucose level of group B was significantly ($P \leq 0.002$) than that of *C. verum* treated group (C) and the control group (A), also the glucose level of *C. verum*

Table (3): The effect of oral administration of *C. verum* and Glibenclamide on serum glucose level of an induced hyperglycemic rats.

Sampling Time	Blood glucose concentration (mg/ dl) of groups:			P
	A	B	C	
0 h	91.57±8.32 [a]	89.52±6.98 [a]	111.17±14.05 [a]	0.2868
1 h	208.58±23.85 [a]	92.49±7.00 [b]	242.08±30.21 [a]	0.0007
2 h	185.83±15.73 [a]	78.05±5.61 [b]	109.25±23.42 [b]	0.0011
4 h	142.67±15.50 [a]	74.92±5.47 [b]	125.25±11.37 [a]	0.0024

(Data are expressed in mean ± SE)

Mean values having different superscript small letters within same row differ significantly at ($P \leq 0.05$).

Group A: Hyperglycemic untreated rats.

Group B: Hyperglycemic treated with Glibenclamide.

Group C: Hyperglycemic treated with *C. verum*.

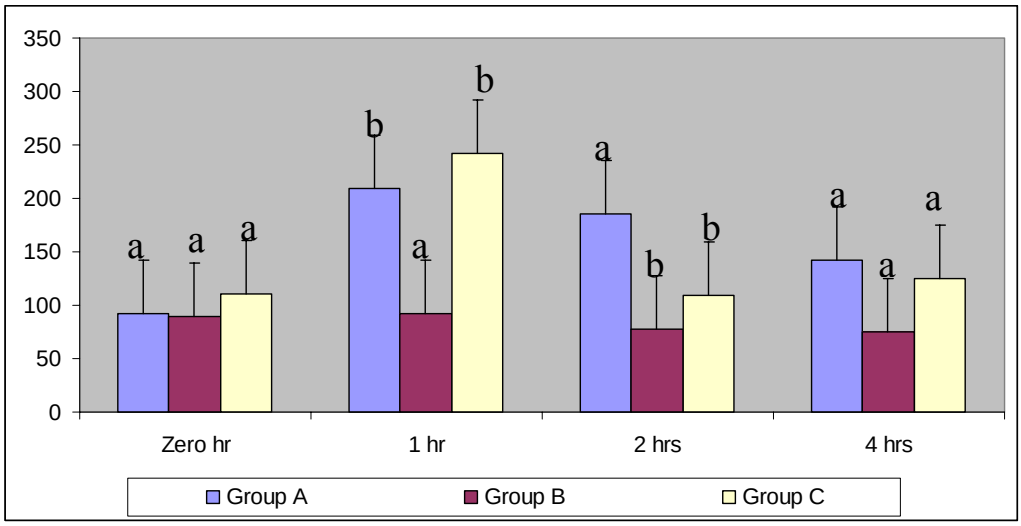

Fig. 6: The effect of *C. verum* and Glibenclamide on serum glucose level of hyperglycemic rats as shown by glucose tolerance test.
Bars with different letters are significantly different ($P \leq 0.05$).

Group A: Hyperglycemic untreated rats.

Group B: Hyperglycemic treated with Glibenclamide.

Group C: Hyperglycemic treated with *C. verum*.

treated group C was nonsignificantly lower than the glucose of the control group A (125.67 Vs 125.25) as shown in Table (3) and Fig. (6).

Experiment 2

3.2 The effects of oral administration of *C. verum* on serum glucose level of normoglycemic rats:

When the serum glucose level of normoglycemic fasting rats was measured after 18 hours fasting (0h) and then at 1, 2 and 4 hours after administration of *C. verum* at a dose of 1g/kgBwt to group E rats, the serum glucose level of the control group (D) and treated group (E) are shown in Table (4) Fig. (7).

Results showed that there was a decrease non-significantly in serum glucose concentration at hour1and 2 in group E when compared to the control group (D).

However, the serum glucose level of *C. verum* treated rats (group E) was significantly lower when compared to control at the hour 4.

It was also observed that the serum glucose level of *C. verum* treated rats decreased gradually after administration of *C. verum* while the serum glucose level of the control remained almost constant throughout the experiment.

Table (4): The effects of administration of *C.verum* on serum glucose level of normoglycemic rats.

Sampling Time	Serum glucose concentration (mg/ dl) of group:		P
	D	E	
0 h	90.36±1.88[a]	95.64±13.92[a]	0.73
1 h	95.59±7.67[a]	88.92±4.62[a]	0.66
2 h	97.03±4.34[a]	80.10±7.33[a]	0.09
4 h	96.56±4.37[a]	73.65±5.08[b]	0.01

(Data are expressed in mean ± SE)

Mean values having different superscript small letters within same row differ significantly at ($P \leq 0.05$).

Group D: Normoglycemic untreated rats.

Group E: Normoglycemic treated with *C. verum*.

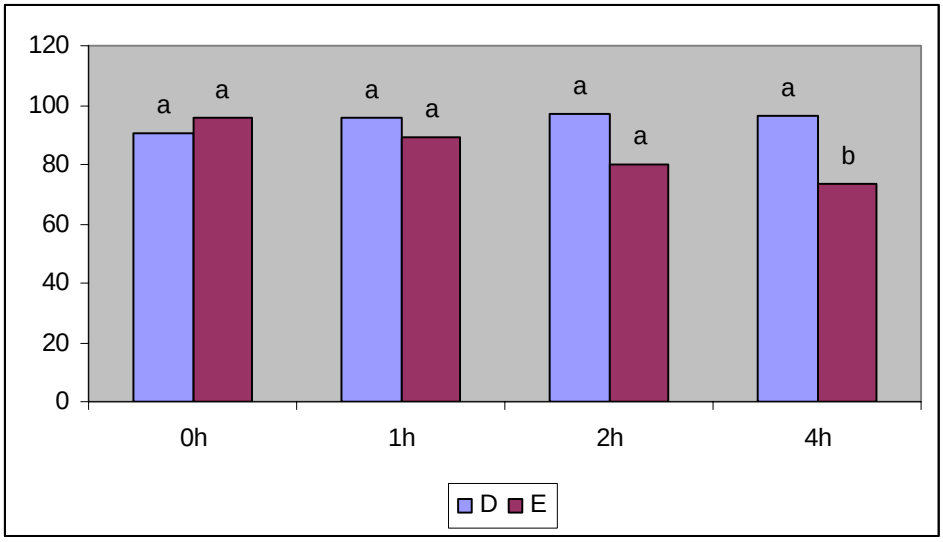

Fig. 7: The effects of administration of *C.verum* on serum glucose level of normoglycemic rats.
Bars with different letters are significantly different at P≤0.05.

Group D: Normoglycemic untreated rats.

Group E: Normoglycemic treated with *C. verum*.

CHAPTER FOUR

DISCUSSION

The current study was carried out to investigate the putative hypoglycemic and antidiabetic effect of *Cinnamomum verum*, one of Sudanese flavouring agents. *C. verum* is one of the tropical plants related to family Lauraceae. The medicinal using of Cinnamon as antidiabetic agent, was investigated by Khan *et al*. (2003), a pilot clinical trial was conducted in Pakistan to study the effect of *Cinnamon cassia* powder in patients with type 2 diabetes, average age was 52.2 ± 6.32. The study showed, successful result, that *Cinnamomum cassia* reduced fasting serumglucose (18 – 29%). *Cinnamomum cassia* a species closely related to *Cinnamomum verum* ("Cassia." Wikipedia, 2007). In this study, *C. verum* was examined for the hypoglycemic effect in induced hyperglycemic rats and normoglycemic fasting rats.

Experiment 1 was conducted to study the effect of *C. verum* bark powder and Gibenclamide in glucose loaded rats. The study showed that *C. verum* powder reduced significantly serum glucose level at 2h and non-significantly at 4h when compared to the control but the reduction in serum glucose was less when compared to the antidiabetic drug Glibenclamide which caused significant reduction at 1, 2 and 4 hours when compared to control. These results are similar those Khan *et al*. (2007); however the *C. verum* in this experiment was administrated to the rats only at 0 h after fasting and not before. The results of this experiment are also in line with Mang *et al*. (2007) who reported moderate glucose lowering effect of aqueous cinnamon extract on fasting plasma glucose

(10.3% Vs 3.37%) when compared to the control group of type 2 diabetes. However, the hypoglycemic effect observed in the present study and previous ones (Khan *et al*., 2003; Mang *et al*., 2007) was not noticed by Vanschoonbeek *et al*. (2006) who found no significant difference in the levels of fasting plasma glucose, and insulin at two weeks and six weeks of treatment. The results of this study also showed that *C. verum* produced hypoglycemic effect but to lesser extend when compared to the commonly used antidiabetic drug Glibenclamide.

Experiment 2 was conducted to study the effect of oral administration of *C. verum* in normoglycemic fasting rats. The results of this experiment showed *C. verum* reduced significantly the serum glucose level at hour 4 following treatment and nonsignificantly at hour 1 and 2 when compared to control group which their glucose level remained almost constant throughout the experimental period. This proved that, *C. verum* could induced reduction of serum glucose level even of fasting animal. Khan *et al*. (2003) investigation revealed that cinnamon reduced blood glucose of type 2 diabetic patients whom received cinnamon for sometime while in experiment 2 of this study *C. verum* first administration reduced the fasting serum glucose level. The experiment 1 of this of investigation showed that *C. verum* also reduced the serum glucose level of glucose loaded rats. These experiments confirm cinnamonhas hypoglycemic effect.

Jarvill-Taylor *et al*. (2001) established that the active component in cinnamon responsible for its insulin-like activity is a water-soluble chemical compound called methylhydroxychalcone polymer (MHCP). They found that MHCP was highly effective, providing essentially the

same biological activity as insulin itself. It was effective not only in increasing the uptake of glucose by cells, but also of stimulating the synthesis of glycogen, a polymeric form of glucose that is stored primarily in the liver and muscle tissues for use at times of peak energy demand, such as exercise. MHCP turned out to be synergistic with insulin in these actions, providing a net effect greater than the sum of its parts. MHCP makes cells more responsive to insulin, i.e., it increases insulin sensitivity, the opposite of insulin resistance. The *C. verum* used in the present experiments was not an extract of *C. verum* bark but rather whole powdered bark which of course contains MHCP along with every other compound of the bark, water soluble or not, thus the observed hypoglycemic of *C. verum* in this study may be attributed to its MHCP content. Researchers in Japan found recently that when an aqueous extract of cinnamon (containing MHCP, of course) was given orally to laboratory rats, the insulin receptors on skeletal muscle cells became more responsive (Qin *et al*., 2003). Enhanced insulin sensitivity means more glucose going into the cells, so the blood glucose levels fall, and biochemical order is restored. By enhancing insulin signaling, cinnamon can prevent insulin resistance even in animals fed a high-fructose diet. Qin *et al*. (2004) showed that when rats fed a high-fructose diet were also given cinnamon extract, their ability to respond to and utilize glucose was improved so much that it was the same as that of rats on a normal (control) diet.

Both test tube and animal studies have shown that compounds in cinnamon not only stimulate insulin receptors, but also inhibit an enzyme that inactivates them, thus significantly increasing cells' ability to use

glucose (Anderson *et al*., 2003). Anderson *et al*. (2003) characterized the insulin-enhancing complexes in cinnamona collection of catechin/ epicatechin oligomers that increase the body's insulin-dependent abiltiy to use glucose roughly 20-fold.

Antioxidant are essential to human body to neutralize free-reactive oxygen radical, to maintain functional cellular membrane and structure. Furthermore free radicals associated with impaired glucose metabolism and antioxidants have been implicated in regression of diabetes mellitus. Cinnamon contains oligomeric proanthocyanidins (OPC) which has anti-oxidantive effect (Gu *et al*., 2003). The observed hypoglycemic effect of *C. verum* in this study may also be attributed to its content of OPC.

When rats were given a daily dose of cinnamon (300 mg Kg Bwt) for a three week period, their skeletal muscle was able to absorb 17% more blood sugar per minute compared to that of control rats, which had not received cinnamon. Khan *et al*. (2003) concluded that, including cinnamon in the diet of people with type 2 diabetes will reduce risk factors associated with diabetes and cardiovascular diseases. However, some scientists had been concerned about potentially toxic effects of regularly consuming cinnamon. New research showed that the potentially toxic compounds in cinnamon bark are found primarily in the lipid soluble fractions and are present only at very low levels in water soluble cinnamon extracts, which are the ones with the insulin-enhancing compounds. Cinnamon does not contain a measurable amount of goitrogen, oxalate or purine and is not known to cause food allergies (WH Food, 2007). Hence it could be concluded cinnamon water extract

can be used as hypoglycemic agent unless otherwise, further study proved it is toxic.

CONCLUSIONS AND RECOMMENDATIONS

Conclusions:

From observations and results of this investigation, it can be concluded that:

1. *C. verum* bark powder decrease significantly plasma glucose level in the induced hypergycemic rats.
2. Therefore, *C. verum* could be considered a promising antidiabetic agent which can be used to reduce blood glucose in diabetic patients.
3. The effect of *C. verum* in the dose 1g/ kg body weight was less, when was compared to the effect of 10 mg/kgBwt of Gibenclamide.
4. The dose 1g/kgBwt of *C. verum* bark powder has hypoglycemic effect in normoglycemic fasting rats.

Recommendation:

Since *Cinnamomum verum* is promising antidiabetic agent it is recommended that:

1. The active ingredient in *C. verum* should be identified and fractionated.
2. Future research should be carried out to determine the mechanism of action of each fraction on blood glucose level.
3. The proper dosage and potential interactions with other medicinal plants and synthetic drugs, has to be studied.

4. The safety of *C. verum* should be examined by administrating very high dose for long period of time.
5. The effect of *C. verum* to type 1diabetes has to be examined.

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